SCIENCE MONOGRAPH 19

JUNE 1970

Quality of Alfalfa Hay as Influenced by Field Application of Organophosphorus Insecticides

AGRICULTURAL EXPERIMENT STATION

UNIVERSITY OF WYOMING LARAMIE

Quality of Alfalfa Hay as Influenced by Field Application of Organophosphorus Insecticides[1, 2]

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Introduction

Alfalfa *(Medicago sativa* L.) is an important forage crop in Wyoming. According to Powell (1968), approximately two-thirds of the total hay produced in the state is alfalfa hay. Organophosphorus insecticides are widely used to control economically important insect pests of alfalfa. Little is known, however, concerning the direct influence of these compounds on the quality of alfalfa hay, independent of the insecticidal properties of the materials.

The study reported in this paper determined if 12 organophosphorus compounds applied to field-grown alfalfa modified the chemical composition of alfalfa hay. Such information will be of value in establishing insecticide recommendations for alfalfa.

Organophosphorus materials could be selected not only for their effectiveness as insecticides but also for the favorable or unfavorable effects they may have on the quality of alfalfa hay.

Review of Previous Work

Changes in the chemical composition of plants, other than alfalfa, upon treatment with organophosphorus insecticides have been published. The metabolic response of the plant to phosphate insecticides varies with the insecticide. Moreover, different plants may exhibit different physiological responses to the same insecticide. When applied to tomatoes at three times the recommended rate, parathion caused production of polylobed leaves in contrast to the 3 to 5-lobed normal leaves (McIlrath, 1950).

McIlrath also noted that HETP (hexaethyl tetraphosphate) and tepp (tetraethyl pyrophosphate) applied at 15 to 30 times the recommended dose, caused 2,4-D (2,4-dichlorophenoxyacetic acid)-like stimulation of cell growth in tomatoes.

Hall (1950) treated carnations and tomatoes with HETP and tepp. The carnations showed total phosphorus increased and total nitrogen decreased. Low concentrations (400 ppm or less) of tepp stimulated growth of flowering tomato plants while concentrations of 800-1,000 ppm inhibited growth. Stimulatory concentrations involved oxygen uptake and decreased dry weight and reserve carbohydrates. A rise in soluble sugars occurred as the reserve carbohydrates were hydrolyzed. At the same time the total phosphorus and nitrogen level increased.

Schradan affected various chemical fractions in cotton. The level of chlorophyll and carotenoid pigments and dry weight of whole plants attained a peak at 10 ppm of schradan, then dropped at higher concentrations. Total sugars, however, peaked at 100 ppm, then decreased, whereas starch, total nitrogen and phosphorus content of the whole plant increased with an increase in concentration of insecticide (Hacskaylo and Ergle, 1955).

Probst and Everly (1957) found that field application of demeton and schradan to soybeans had no effect on height, dry weight, protein, or oil content.

Hacskaylo (1957) studied the growth response of cotton grown in sand cultures containing various levels of phorate. The level of reducing

[1] This study was supported by Project 920 from the Entomology Research Division, Agr. Res. Serv., USDA, to the Entomology Section of the Plant Science Division.

[2] Portion of a dissertation presented by the senior author to the Graduate School, University of Wyoming, in partial fulfillment of the requirements for the Ph.D. degree in Entomology.

[3] Formerly, Graduate Research Assistant; Professor of Entomology; and formerly, Professor of Entomology, respectively.

sugars, sucrose, starch, and phosphorus increased, while the dry weight, soluble and protein nitrogen decreased.

Lichtenstein *et al.* (1962) grew corn and pea plants in quartz sand treated with five organophosphorus insecticides—parathion, methyl parathion, phorate, malathion and demeton at a rate of 30 ppm. All these insecticides inhibited corn root growth. Growth of pea roots was depressed only by methyl parathion, phorate and demeton. Parathion increased the respiration rate of corn root tips; demeton reduced the respiration rates of root tips of oats and peas, but not of cucumbers or corn. Dry matter was increased in the roots of corn, oats, peas and cucumbers treated with all five of the insecticides.

The level of total nitrogen and ascorbic acid increased in schradan-treated corn (Alekseev and Izotova, 1962).

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Materials and Methods

Ranger alfalfa, *Medicago sativa* L., was treated with 12 organophosphorus insecticides at the University of Wyoming agronomy farm in Laramie. Weather data (Appendix Table 1) during the course of the 1965-66 studies were recorded at the Federal Aviation Airport, located a few miles north of the agronomy farm. A low-pressure, tractor-mounted sprayer, with 12 nozzles on the boom fixed 40 inches above the ground, was used to apply the compounds. Each insecticide was applied at the rate of 12 gal of total spray per acre at a pressure of 40 psi.

The following insecticides were applied:

1. Malathion [0,0-dimethyl dithiophosphate of diethyl mercaptosuccinate] 5 lbs active ingredient per gal; two pints per acre or 1.25 lbs active ingredient applied per acre.
2. Naled [1,2-dibromo-2,2-dichloroethyl dimethyl phosphate] 8 lbs active ingredient per gal; one pint per acre or 1.0 lb active ingredient applied per acre.
3. Phosphamidon [2-chloro-2-diethylcarbamoyl-l-methylvinyl dimethyl phosphate] 4 lbs active ingredient per gal; one pint per acre or 0.5 lb active ingredient applied per acre.
4. Diazinon [0,0-diethyl 0-(2-isopropyl-6-methyl-4-pyrimidinyl) phosphorothioate] 4 lbs active ingredient per gal; one pint per acre or 0.5 lb active ingredient applied per acre.
5. Dimethoate [0,0-dimethyl S-(N-methylcarbamoylmethyl) phosphorodithioate] 2.67 lbs active ingredient per gal; one pint per acre or 0.33 lb active ingredient applied per acre.
6. Parathion [0,0-diethyl 0-p-nitrophenyl phosphorothioate] 4 lbs active ingredient per gal; one-half pint per acre or 0.25 lbs active ingredient applied per acre.
7. Mevinphos [0,0-dimethyl l-carbomethoxy-l-propen-2-yl phosphate] 2 lbs active ingredient per gal; one-half pint per acre or 0.125 lb active ingredient applied per acre.
8. SD 7438; [S,S/-benzylidene bis-(0,0-dimethyl phosphorodithioate)] 2 lbs active ingredient per gal; one-half pint per acre or 0.125 lb active ingredient per acre.
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10. GS-13005; [S-(2-methoxy-5-oxo-Δ^2-1,3,4-(thiadiazolin-4-yl)-methyl) 0,0-dimethyl) phosphorodithioate] 3.55 lbs active ingredient per gal; two and one-half pints per acre of 1.11 lbs active ingredient applied per acre.
11. Demeton [0,0-diethyl 0 (and S)-2-(ethylthio) ethyl phosphorothioates] 2 lbs active ingredient per gal; one pint per acre or 0.25 lbs active ingredient per acre applied.

12. Trichlorfon [0,0-dimethyl 2,2,2-trichloro-1-hydroxyethyl phosphorate] 50% S.P.; one lb per acre or 0.5 lb active ingredient applied per acre.

Spray applications were made once during the summer of 1965 on the regrowth (second cutting). In 1966, two applications were made, one during the spring growth (first cutting) and one during the summer growth period (second cutting). During both years insecticides were applied when the alfalfa was six-10 inches tall. After the spring cutting of 1966, plots were re-established at another site where they were subjected to the same insecticide treatments.

All plots were arranged in a split plot design with date of sampling as whole plots and insecticides as subplots. The 12 plots treated with insecticides, plus a control plot, were randomized within each of three contiguous blocks. Each plot in the 1965 tests was 11 x 50 feet whereas in 1966, plots were increased to 11 x 100 feet to provide more area for sampling.

To minimize drift, plots were sprayed during early morning when the wind velocity was minimal. To prevent insecticides from being washed off the plants, they were applied on clear days when probability of rainfall was low.

During the 1965 experiment, samples were taken from each plot of alfalfa eight, 14, 21, 28, and 35 days after application. In the 1966 experiments, samples were collected after one, five, seven, 14, 21, and 28 days. Five-pound alfalfa samples were collected from each plot on each sampling date using a power mower. Samples were taken randomly no less than 2 ft from the edges of the plot. Samples then were placed in cloth bags and transferred to a well-ventilated barn where they were spread out and air-dried. Periodically, samples were turned to facilitate drying. When dry, they were ground in a Wiley mill to pass through a 40 mesh screen, were mixed thoroughly with a spatula, and were stored in air-tight pint bottles.

Analytical procedures, as outlined by the Association of Official Agricultural Chemists (1965), were used to determine percent crude protein, crude fiber, ether extract, ash, and total phosphorus. All data are expressed on a dry weight basis. Data were subjected to an analysis of variance for a split plot design (Steel and Torrie, 1960).

Results and Discussion

Appendix Tables 2 through 7 summarize the average percent of five chemical constituents in alfalfa at each sampling date for 12 insecticide treatments. These data failed to reveal any trends resulting from treatment with the insecticides.

Average percent content of each chemical constituent for all sampling dates is presented in Appendix Tables 8 through 13. When the highest and lowest means are carefully examined, differences in the levels of some chemical constituents for insecticide treatments relative to the controls are apparent.

Highest and lowest mean percentages of six chemical constituents in alfalfa according to insecticide treatment are given in Table 14. Some general comments may be made about the most frequently appearing insecticide in the table. Considering the summer growth period of 1965, trichlorfon-treated plots exhibited the lowest percent crude protein, phosphorus, and ether extract but highest percent crude fiber. During the summer growth period of 1966, phosphamidon-treated plots were highest in percent nitrogen-free extract but lowest in percent crude protein, ether extract and ash. Alfalfa treated with malathion was highest in percent ether extract during spring and summer growth in 1966.

Considering both the 1965 and 1966 studies, parathion-treated plants ranked highest in percent crude protein during summer growth of both years. During summer growth of 1965 and 1966 plants treated with phosphamidon were lowest in ash content. Alfalfa treated with azinphosmethyl ranked highest in the following chemical constituents: percent ether extract during summer growth of 1965; percent nitrogen-free extract during spring growth of 1966; and percent phosphorus during summer growth of 1966. Dimethoate-treated alfalfa was highest in percent nitrogen-free extract during summer growth of 1965, and highest in crude fiber and ash during summer growth of 1966.

On the other hand, treated alfalfa which ranked highest in percent of certain chemical constituents in one growth period ranked lowest for the same constituent in the next growth period. Percent crude protein content of phosphamidon treated alfalfa was highest during spring growth but lowest during summer growth of 1966. Dimethoate-treated plants were lowest in percent crude fiber during summer growth of 1965 but highest during the same growth period of 1966.

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Although some of the insecticides evaluated in this study appeared to alter the levels of certain chemical constituents in alfalfa, none of the treatments was statistically significant. The analysis of variance of percent crude protein, crude fiber, ether extract, nitrogen-free extract, phosphorus, and ash for summer growth of 1965 and spring and summer growth of 1966 revealed that none of the F ratios for whole plots (insecticides) or whole plots (insecticides) X subplots (sampling dates) was significant at the 0.05% level (Appendix Tables 15 through 20). F ratios for subplots (sampling dates) were highly significant in both the 1965 and 1966 studies. Since changes in the levels of certain chemical constituents in the controls would normally be expected with advance in maturity, these means were not tested for significant differences.

In general, 12 organophosphate insecticides at the rates they were applied in this study did not significantly alter the chemical composition of alfalfa hay during the summer growth of 1965 or spring and summer growth of 1966.

Summary

The effect of organophosphorus insecticides on certain chemical constituents in alfalfa was studied. Experiments were conducted in the field at the University of Wyoming agronomy farm in Laramie. Twelve organophosphorus insecticides were applied during summer growth of 1965 and spring and summer growth of 1966. Samples were collected periodically as alfalfa matured, and the following chemical determinations were made: crude protein, crude fiber, ether extract, nitrogen-free extract, ash, and phosphorus.

Changes in the levels of certain chemical constituents occurred in alfalfa treated with trichlorfon, phosphamidon, parathion, malathion, azinphosmethyl, and dimethoate. However, an analysis of variance of percent crude protein, crude fiber, ether extract, nitrogen-free extract, ash, and phosphorus in alfalfa indicated that none of the insecticide treatments was significantly different at the 0.05% level for summer growth of 1965 or spring and summer growth of 1966. These data, therefore, indicated that 12 organophosphate insecticides at the rates evaluated in this study may be used to control insect pests on alfalfa without significantly affecting the quality of alfalfa hay.

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Appendix

Table 1. Precipitation and temperature conditions that prevailed during the summers of the 1965 and 1966 Studies[a].

	1965 Study				1966 Study			
	Temperature F°							
Month	Mean minimum	Mean maximum	Average	Departure from normal[b]	Mean minimum	Mean maximum	Average	Departure from normal[b]
May	31.9	58.8	45.4	−3.40	32.1	67.5	49.8	+1.20
June	42.1	69.0	55.6	−1.22	41.2	72.8	57.0	+0.32
July	47.6	79.0	63.3	−0.84	50.8	84.7	67.8	+3.42
August	43.7	75.7	59.7	−2.70	44.4	77.5	61.0	−0.84
	Precipitation							
Month	Total inches	Departure from normal[b]	Greatest day	Date	Total inches	Departure from normal[b]	Greatest day	Date
May	2.62	+1.00	0.75	14	0.01	−1.63	0.01	22
June	1.63	+0.21	0.41	10	1.43	+0.09	0.44	8
July	2.02	+0.35	0.78	25	1.30	−0.160	0.51	21
August	0.41	−0.11	0.18	13	0.92	−0.01	0.47	2

[a]The precipitation and temperature were recorded at the Federal Aviation Airport located approximately two miles north of the alfalfa plots. In Climatological Data. 1964-65. U. S. Department of Commerce. 74 (5-8): 71-130; 75 (5-8): 67-119.

[b]Departure from normal based on weather data for previous five years.

Table 2. Average percent crude protein in alfalfa sampled at various dates after application of 12 organophosphorus insecticides during summer growth of 1965 and spring and summer growth of 1966.

Date sampled	No. of days after application of insecticide	% crude protein						
		Malathion	Naled	Phosphamidon	Diazinon	Dimethoate	Parathion	Mevinphos
					1965 Study			
					Summer growth			
8/5	8	23.34	23.98	23.43	22.95	23.57	24.72	23.98
8/11	14	23.32	23.73	24.27	24.23	24.34	24.41	23.49
8/18	21	23.93	22.85	22.00	22.26	23.02	23.63	22.67
8/25	28	21.77	22.48	21.99	22.42	22.66	22.30	22.61
9/1	35	19.96	19.79	19.83	20.26	20.52	20.36	19.75
					1966 Study			
					Spring growth[a]			
6/18	1	20.17	20.90	20.59	20.20		20.91	19.88
6/22	5	23.42	22.28	22.91	22.77		22.46	21.54
6/24	7	23.11	23.28	23.65	23.40		22.64	23.53
7/1	14	24.11	24.19	25.44	24.25		23.35	23.34
7/8	21	24.73	24.77	24.74	24.97		25.62	24.55
					Summer growth			
7/30	1	29.14	29.31	28.66	28.82	29.04	29.78	28.94
8/3	5	25.07	25.19	25.15	26.75	25.59	25.49	25.59
8/5	7	26.35	26.43	26.39	26.84	27.28	27.18	26.54
8/12	14	24.29	24.75	24.86	25.49	25.41	26.24	25.48
8/19	21	24.54	24.59	24.04	25.17	23.95	24.57	25.19
8/26	28	23.06	22.41	21.73	22.05	22.33	22.66	21.99

Date sampled	No. of days after application of insecticide	% crude protein					
		S D 7438	Azinphosmethyl	G S 13005	Demeton	Trichlorfon	Control
				1965 Study			
				Summer growth			
8/5	8	24.14	24.46	23.84	21.83	22.87	21.83
8/11	14	23.62	24.17	24.07	24.71	23.89	24.71
8/18	21	22.31	23.56	22.04	22.31	22.77	22.32
8/25	28	21.76	22.08	22.30	21.86	21.26	21.86
9/1	35	20.06	20.07	19.92	20.04	19.54	20.04
				1966 Study			
				Spring growth[b]			
6/18	1	20.91	21.47	20.25			22.28
6/22	5	22.91	22.36	22.45			22.98
6/24	7	23.23	23.39	22.94			23.50
7/1	14	24.08	22.87	23.99			24.52
7/18	21	25.41	24.33	25.60			25.30
				Summer growth			
7/30	1	28.90	28.81	29.78	28.92	28.80	29.00
8/3	5	25.72	25.29	25.15	25.43	24.28	24.22
8/5	7	26.92	27.54	27.58	26.90	27.66	27.48
8/12	14	25.37	24.74	25.47	25.26	25.63	25.09
8/19	21	24.56	25.36	24.82	24.65	25.02	24.56
8/26	28	22.88	22.86	22.27	21.76	22.90	22.17

[a] Dimethoate was not applied during spring stage of growth.
[b] Demeton and trichlorfon were not applied during spring stage of growth.

Table 3. Average percent crude fiber in alfalfa sampled at various dates after application of 12 organophosphorus insecticides during summer growth of 1965 and spring and summer growth of 1966.

Date sampled	No. of days after application of insecticide	% crude fiber						
		Malathion	Naled	Phosphamidon	Diazinon	Dimethoate	Parathion	Mevinphos
					1965 Study			
					Summer growth			
8/5	8	21.38	21.18	21.11	21.29	21.67	21.22	21.62
8/11	14	22.78	22.13	21.99	22.03	19.71	22.80	22.13
8/18	21	23.89	24.46	26.80	25.27	23.69	24.79	25.02
8/25	28	25.71	24.69	25.41	24.86	24.20	25.11	24.80
9/1	35	26.89	25.88	26.66	25.99	25.66	26.11	25.85
					1966 Study			
					Spring growth[a]			
6/18	1	22.38	23.70	22.65	25.22		23.90	23.42
6/22	5	25.52	25.74	24.62	25.12		24.71	26.28
6/24	7	24.82	25.85	23.72	25.57		25.90	25.87
7/1	14	25.34	24.60	24.19	25.38		25.16	26.37
7/8	21	24.69	25.54	24.77	25.27		24.77	25.90
					Summer growth			
7/30	1	22.27	22.10	22.12	21.62	21.42	21.01	21.61
8/3	5	25.42	25.56	24.64	23.68	25.58	26.50	25.33
8/5	7	23.93	25.05	24.28	25.11	24.09	23.71	23.80
8/12	14	24.97	24.57	24.58	23.95	23.77	23.55	25.03
8/19	21	23.97	24.18	25.35	25.15	26.76	25.44	24.90
8/26	28	20.95	21.90	22.32	21.68	23.08	22.64	22.40

Date sampled	No. of days after application of insecticide	% crude fiber					
		S D 7438	Azinphosmethyl	G S 13005	Demeton	Trichlorfon	Control
				1965 Study			
				Summer growth			
8/5	8	21.19	20.92	21.31	22.52	24.47	24.03
8/11	14	21.84	22.04	22.01	21.74	22.31	21.37
8/18	21	25.01	24.18	25.09	24.97	25.47	23.71
8/25	28	26.11	24.19	24.93	25.19	26.56	25.66
9/1	35	24.57	27.65	27.00	26.68	27.14	26.91
				1966 Study			
				Spring growth[b]			
6/18	1	24.22	23.93	23.19			22.37
6/22	5	26.65	23.93	25.22			25.15
6/24	7	25.97	25.03	25.33			24.96
7/1	14	25.92	25.01	25.53			24.50
7/8	21	25.22	25.67	24.42			25.36
				Summer growth			
7/30	1	21.76	21.82	21.52	21.55	21.79	21.80
8/3	5	25.61	24.81	24.48	24.68	25.41	24.52
8/5	7	23.23	23.89	23.77	23.69	24.52	23.88
8/12	14	24.42	24.03	23.16	23.79	23.79	24.34
8/19	21	24.19	24.76	25.68	25.25	25.29	25.16
8/26	28	20.91	21.38	22.32	23.21	21.84	22.41

[a] Dimethoate was not applied during the spring stage of growth.
[b] Demeton and trichlorfon were not applied during the spring stage of growth.

Table 4. Average percent ether extract in alfalfa sampled at various dates after application of 12 organophosphorus insecticides during summer growth of 1965 and spring and summer growth of 1966.

Date sampled	No. of days after application of insecticide	% ether extract						
		Malathion	Naled	Phosphamidon	Diazinon	Dimethoate	Parathion	Mevinphos
					1965 Study			
					Summer growth			
8/5	8	1.91	2.00	1.81	1.85	1.94	1.98	1.96
8/11	14	2.29	2.12	2.15	2.21	1.90	2.29	1.99
8/18	21	1.72	1.85	1.80	1.92	1.89	1.88	2.06
8/25	28	1.92	2.05	1.95	2.22	2.12	1.95	2.19
9/1	35	1.92	1.88	1.96	1.94	2.00	1.85	1.88
					1966 Study			
					Spring growth[a]			
6/18	1	1.32	1.19	1.35	1.45		1.21	1.14
6/22	5	1.24	1.09	1.14	1.10		1.07	1.08
6/24	7	1.38	1.26	1.30	1.25		1.24	1.30
7/1	14	1.50	1.40	1.50	1.45		1.34	1.32
7/8	21	1.40	1.44	1.44	1.44		1.46	1.54
					Summer growth			
7/30	1	1.43	1.49	1.34	1.45	1.51	1.61	1.49
8/3	5	1.47	1.57	1.42	1.54	1.41	1.36	1.48
8/5	7	1.62	1.37	1.37	1.40	1.39	1.56	1.46
8/12	14	1.50	1.24	1.42	1.53	1.58	1.61	1.56
8/19	21	1.38	1.22	1.24	1.32	1.18	1.27	1.30
8/26	28	1.66	1.45	1.51	1.47	1.31	1.48	1.43

Date sampled	No. of days after application of insecticide	% ether extract					
		S D 7438	Azinphosmethyl	G S 13005	Demeton	Trichlorfon	Control
				1965 Study			
				Summer growth			
8/5	8	1.98	2.04	1.82	1.77	1.81	1.86
8/11	14	2.29	2.39	2.13	2.16	2.06	2.11
8/18	21	1.96	1.83	1.96	1.76	1.76	1.95
8/25	28	2.09	2.34	2.15	2.18	1.92	2.07
9/1	35	2.03	1.85	1.85	1.70	1.49	1.95
				1966 Study			
				Spring growth[b]			
6/18	1	1.01	1.27	1.26			1.06
6/22	5	0.98	1.15	1.19			1.07
6/24	7	1.25	1.32	1.36			1.33
7/1	14	1.37	1.37	1.48			1.30
7/8	21	1.49	1.41	1.46			1.49
				Summer growth			
7/30	1	1.54	1.46	1.42	1.52	1.57	1.47
8/3	5	1.37	1.57	1.46	1.46	1.50	1.44
8/5	7	1.50	1.34	1.35	1.36	1.38	1.38
8/12	14	1.51	1.60	1.60	1.61	1.50	1.40
8/19	21	1.35	1.37	1.46	1.34	1.37	1.30
8/26	28	1.51	1.46	1.43	1.42	1.45	1.59

[a] Dimethoate was not applied during spring stage of growth.
[b] Demeton and trichlorfon were not applied during spring stage of growth.

Table 5. Average percent nitrogen-free extract in alfalfa sampled at various dates after application of 12 organophosphorus insecticides during summer growth of 1965 and spring and summer growth of 1966.

Date sampled	No. of days after application of insecticide	% nitrogen-free extract						
		Malathion	Naled	Phosphamidon	Diazinon	Dimethoate	Parathion	Mevinphos
					1965 Study			
					Summer growth			
8/5	8	38.08	37.96	38.18	38.77	37.93	37.52	38.04
8/11	14	37.31	38.36	38.09	37.26	39.05	36.31	37.67
8/18	21	36.29	36.67	36.67	36.56	37.04	35.58	36.66
8/25	28	38.71	38.88	38.69	37.86	38.76	38.50	37.81
9/1	35	39.73	40.10	39.87	40.06	40.40	40.14	40.47
					1966 Study			
					Spring growth[a]			
6/18	1	38.88	37.42	39.83	37.71		38.36	38.91
6/22	5	35.70	36.99	37.13	37.18		38.47	37.87
6/29	7	36.10	35.55	36.85	34.32		36.12	33.85
7/1	14	33.81	37.20	33.82	34.74		36.55	36.63
7/8	21	35.74	34.34	36.29	35.99		36.26	34.32
					Summer growth			
7/30	1	30.84	30.86	32.59	32.83	32.64	31.14	32.68
8/3	5	32.75	31.14	34.05	32.93	31.28	31.77	32.46
8/5	7	32.87	33.21	33.11	31.36	31.70	31.73	32.97
8/12	14	33.03	34.13	32.83	33.32	30.98	31.94	31.42
8/19	21	35.85	36.25	36.28	34.33	34.33	34.85	33.90
8/26	28	40.96	40.89	40.72	41.61	40.32	39.52	40.31

Date sampled	No. of days after application of insecticide	% nitrogen-free extract					
		S D 7438	Azinphosmethyl	G S 13005	Demeton	Trichlorfon	Control
				1965 Study			
				Summer growth			
8/5	8	38.39	37.93	38.13	37.20	31.19	37.77
8/11	14	37.93	37.01	37.92	36.92	37.52	37.69
8/18	21	37.34	35.88	36.60	35.99	35.82	37.59
8/25	28	37.80	38.50	37.72	37.55	37.81	38.04
9/1	35	41.39	39.81	40.25	39.88	40.22	40.50
				1966 Study			
				Spring growth[b]			
6/18	1	38.83	38.47	39.27			39.40
6/22	5	36.76	38.93	37.36			36.32
6/24	7	34.16	36.21	35.85			34.96
7/1	14	33.63	37.59	34.68			35.81
7/8	21	34.74	34.53	33.73			33.23
				Summer growth			
7/30	1	32.27	31.60	31.90	31.86	31.97	32.69
8/3	5	30.18	31.61	33.46	34.53	33.68	36.02
8/5	7	32.96	32.74	32.52	32.81	31.29	31.54
8/12	14	32.60	32.94	33.81	32.65	32.97	32.11
8/19	21	35.71	34.75	34.04	34.97	34.18	35.23
8/26	28	40.06	41.69	40.56	40.71	40.55	40.04

[a] Dimethoate was not applied during spring stage of growth.
[b] Demeton and trichlorfon were not applied during spring stage of growth.

Table 6. Average percent phosphorus in alfalfa sampled at various dates after application of 12 organophosphorus insecticides during summer growth of 1965 and spring and summer growth of 1966.

Date sampled	No. of days after application of insecticide	% phosphorus						
		Malathion	Naled	Phosphamidon	Diazinon	Dimethoate	Parathion	Mevinphos
					1965 Study			
					Summer growth			
8/5	8	0.351	0.378	0.341	0.361	0.352	0.366	0.366
8/11	14	0.308	0.321	0.323	0.342	0.326	0.322	0.334
8/18	21	0.316	0.327	0.286	0.305	0.298	0.321	0.313
8/25	28	0.264	0.281	0.277	0.293	0.278	0.280	0.299
9/1	35	0.242	0.252	0.249	0.260	0.248	0.256	0.266
					1966 Study			
					Spring growth[a]			
6/18	1	0.237	0.254	0.253	0.250		0.233	0.255
6/22	5	0.279	0.280	0.272	0.304		0.267	0.282
6/24	7	0.279	0.294	0.306	0.321		0.283	0.329
7/1	14	0.293	0.280	0.285	0.293		0.255	0.286
7/8	21	0.286	0.303	0.289	0.298		0.289	0.313
					Summer growth			
7/30	1	0.373	0.397	0.350	0.332	0.352	0.385	0.342
8/3	5	0.248	0.271	0.259	0.297	0.256	0.285	0.257
8/5	7	0.258	0.303	0.310	0.326	0.341	0.275	0.304
8/12	14	0.224	0.258	0.262	0.251	0.240	0.247	0.244
8/19	21	0.248	0.256	0.233	0.258	0.245	0.248	0.212
8/26	28	0.191	0.173	0.173	0.173	0.206	0.195	0.176

Date sampled	No. of days after application of insecticide	% phosphorus					
		S D 7438	Azinphosmethyl	G S 13005	Demeton	Trichlorfon	Control
				1965 Study			
				Summer growth			
8/5	8	0.356	0.359	0.355	0.350	0.342	0.329
8/11	14	0.334	0.326	0.318	0.334	0.315	0.320
8/18	21	0.292	0.299	0.285	0.302	0.303	0.290
8/25	28	0.270	0.266	0.273	0.276	0.261	0.269
9/1	35	0.248	0.239	0.243	0.247	0.239	0.237
				1966 Study			
				Spring growth[b]			
6/18	1	0.247	0.242	0.246			0.256
6/22	5	0.291	0.269	0.280			0.284
6/24	7	0.292	0.274	0.289			0.315
7/1	14	0.281	0.255	0.290			0.291
7/8	21	0.309	0.282	0.308			0.311
				Summer growth			
7/30	1	0.338	0.364	0.359	0.367	0.339	0.356
8/3	5	0.233	0.233	0.272	0.304	0.241	0.324
8/5	7	0.271	0.336	0.310	0.309	0.319	0.274
8/12	14	0.246	0.273	0.238	0.230	0.265	0.255
8/19	21	0.253	0.277	0.264	0.240	0.241	0.248
8/26	28	0.223	0.193	0.182	0.211	0.218	0.172

[a] Dimethoate was not applied during spring stage of growth.
[b] Demeton and trichlorfon were not applied during spring stage of growth.

Table 7. Average percent ash in alfalfa sampled at various dates after application of 12 organophosphorus insecticides during summer growth of 1965 and spring and summer growth of 1966.

Date sampled	No. of days after application of insecticide	% ash						
		Malathion	Naled	Phosphamidon	Diazinon	Dimethoate	Parathion	Mevinphos
					1965 Study			
					Summer growth			
8/5	8	11.16	10.51	10.23	10.64	10.63	10.33	9.92
8/11	14	10.06	9.45	9.48	9.96	10.73	10.05	10.60
8/18	21	9.59	9.81	9.22	9.53	9.92	9.62	9.33
8/25	28	7.83	8.00	7.80	8.57	8.43	8.02	8.66
9/1	35	7.74	8.08	7.56	7.54	7.50	7.47	7.94
					1966 Study			
					Spring growth[a]			
6/18	1	12.00	11.71	10.89	11.18		11.30	12.27
6/22	5	9.12	8.76	9.08	9.30		8.68	8.88
6/24	7	9.63	9.18	9.95	9.67		8.86	9.62
7/1	14	9.89	9.07	9.20	9.22		8.93	10.55
7/8	21	9.58	9.85	9.59	9.10		9.09	9.04
					Summer growth			
7/30	1	11.34	10.89	10.29	10.45	10.61	11.23	10.42
8/3	5	10.48	10.63	9.83	9.87	10.66	10.23	9.96
8/5	7	9.50	9.47	10.09	10.49	10.90	10.27	10.29
8/12	14	10.42	10.55	10.66	9.77	12.10	10.58	10.54
8/19	21	9.88	9.30	8.39	9.39	9.13	9.10	9.77
8/26	28	8.72	9.11	9.24	8.80	8.53	9.30	9.44

Date sampled	No. of days after application of insecticide	% ash					
		S D 7438	Azinphosmethyl	G S 13005	Demeton	Trichlorfon	Control
				1965 Study			
				Summer growth			
8/5	8	9.69	10.18	10.50	12.48	10.34	10.43
8/11	14	10.10	10.23	9.63	10.30	9.97	9.45
8/18	21	9.00	10.04	10.19	10.31	9.84	9.86
8/25	28	8.06	8.36	8.98	9.14	9.40	8.30
9/1	35	7.61	7.02	7.15	7.49	7.33	7.01
				1966 Study			
				Spring growth[b]			
6/18	1	11.27	10.06	10.43			11.20
6/22	5	8.89	9.27	9.27			9.19
6/24	7	9.71	9.21	9.21			9.58
7/1	14	8.88	9.04	9.04			9.28
7/8	21	9.42	9.97	9.97			9.92
				Summer growth			
7/30	1	10.40	11.05	11.06	10.94	10.84	10.88
8/3	5	10.66	10.62	10.08	9.64	9.63	9.44
8/5	7	10.23	9.81	10.00	10.34	10.37	10.27
8/12	14	10.62	10.68	10.04	10.62	10.62	11.47
8/19	21	9.85	9.23	9.17	9.35	9.71	9.18
8/26	28	9.52	8.42	9.06	8.08	8.64	9.21

[a] Dimethoate was not applied during spring stage of growth.
[b] Demeton and trichlorfon were not applied during spring stage of growth.

Table 8. Average percent crude protein in alfalfa over all sampling dates after application of 12 organophosphorus insecticides during summer growth of 1965 and spring and summer growth of 1966.

Insecticide	% crude protein		
	1965 Study Stage of growth	1966 Study Stage of growth	
	Summer	Spring[a]	Summer
Malathion	22.46	23.11	25.41
Naled	22.57	23.08	25.45
Phosphamidon	22.31	23.47	25.14
Diazinon	22.42	23.12	25.84
Dimethoate	22.82	–	25.60
Parathion	23.09	22.99	25.99
Mevinphos	22.50	22.57	25.62
S D 7438	22.38	23.31	25.73
Azinphosmethyl	22.87	22.88	25.77
G S 13005	22.43	23.05	25.85
Demeton	22.15	–	25.49
Trichlorfon	22.07	–	25.71
Control	22.15	23.71	25.42

[a] Dimethoate, demeton and trichlorfon were not applied during spring stage of growth.

Table 9. Average percent crude fiber in alfalfa over all sampling dates after application of 12 organophosphorus insecticides during summer growth of 1965 and spring and summer growth of 1966.

Insecticide	% crude fiber		
	1965 Study Stage of growth	1966 Study Stage of growth	
	Summer	Spring[a]	Summer
Malathion	24.13	24.55	23.58
Naled	23.67	25.08	23.89
Phosphamidon	24.45	23.99	23.88
Diazinon	23.89	25.31	23.53
Dimethoate	22.99	–	24.12
Parathion	24.01	24.89	23.81
Mevinphos	23.89	25.57	23.85
S D 7438	23.74	25.60	23.35
Azinphosmethyl	23.94	24.71	23.45
G S 13005	24.07	24.74	23.49
Demeton	24.22	–	23.69
Trichlorfon	25.19	–	23.77
Control	24.34	24.47	23.69

[a] Dimethoate, demeton and trichlorfon were not applied during spring stage of growth.

Table 10. Average percent ether extract in alfalfa over all sampling dates after application of 12 organophosphorus insecticides during summer growth of 1965 and spring and summer growth of 1966.

Insecticide	% ether extract		
	1965 Study Stage of growth	1966 Study Stage of growth	
	Summer	Spring[a]	Summer
Malathion	1.95	1.38	1.51
Naled	1.98	1.27	1.39
Phosphamidon	1.93	1.35	1.38
Diazinon	2.03	1.28	1.45
Dimethoate	1.97	–	1.40
Parathion	1.99	1.26	1.45
Mevinphos	2.02	1.27	1.45
S D 7438	2.07	1.22	1.47
Azinphosmethyl	2.09	1.30	1.47
G S 13005	1.98	1.35	1.45
Demeton	1.91	–	1.45
Trichlorfon	1.90	–	1.46
Control	1.99	1.25	1.43

[a] Dimethoate, demeton and trichlorfon were not applied during spring stage of growth.

Table 11. Average percent nitrogen-free extract in alfalfa over all sampling dates after application of 12 organophosphorus insecticides during summer growth of 1965 and spring and summer growth of 1966.

Insecticide	% nitrogen-free extract		
	1965 Study Stage of growth	1966 Study Stage of growth	
	Summer	Spring[a]	Summer
Malathion	38.02	36.05	34.39
Naled	38.40	36.30	34.41
Phosphamidon	38.30	36.78	34.93
Diazinon	38.10	35.99	34.40
Dimethoate	38.64	–	33.54
Parathion	37.61	36.95	33.49
Mevinphos	38.13	36.32	33.93
S D 7438	38.57	36.02	33.96
Azinphosmethyl	37.83	37.15	34.22
G S 13005	38.12	36.18	34.38
Demeton	37.51	–	34.59
Trichlorfon	37.52	–	34.11
Control	38.32	35.94	34.61

[a] Dimethoate, demeton and trichlorfon were not applied during spring stage of growth.

Table 12. Average percent phosphorus in alfalfa over all sampling dates after application of 12 organophosphorus insecticides during summer growth of 1965 and spring and summer growth of 1966.

Insecticides	% phosphorus		
	1965 Study Stage of growth	1966 Study Stage of growth	
	Summer	Spring[a]	Summer
Malathion	0.296	0.275	0.257
Naled	0.312	0.282	0.276
Phosphamidon	0.295	0.281	0.265
Diazinon	0.312	0.293	0.273
Dimethoate	0.301	–	0.274
Parathion	0.309	0.265	0.273
Mevinphos	0.316	0.293	0.266
S D 7438	0.300	0.284	0.261
Azinphosmethyl	0.298	0.264	0.279
G S 13005	0.295	0.283	0.271
Demeton	0.302	–	0.277
Trichlorfon	0.292	–	0.271
Control	0.289	0.291	0.271

[a] Dimethoate, demeton and trichlorfon were not applied during spring stage of growth.

Table 13. Average percent ash in alfalfa over all sampling dates after application of 12 organophosphorus insecticides during summer growth of 1965 and spring and summer growth of 1966.

Insecticide	% ash		
	1965 Study Stage of growth	1966 Study Stage of growth	
	Summer	Spring[a]	Summer
Malathion	9.28	10.04	10.06
Naled	9.17	9.71	9.99
Phosphamidon	8.86	9.67	9.75
Diazinon	9.25	9.69	9.79
Dimethoate	9.44	–	10.32
Parathion	9.10	9.37	10.12
Mevinphos	9.29	10.07	10.07
S D 7438	8.89	9.64	10.21
Azinphosemethyl	9.17	9.51	9.97
G S 13005	9.29	9.59	9.90
Demeton	9.94	–	9.83
Trichlorfon	9.18	–	9.97
Control	9.11	9.83	10.08

[a] Dimethoate, demeton and trichlorfon were not applied during spring stage of growth.

Table 14. The highest and lowest average percents of six chemical constituents in alfalfa according to insecticide treatment during the summer growth stage of 1965 and the spring and summer growth stages of 1966.

Summer growth of 1965					Spring growth of 1966					Summer growth of 1966				
Highest		Lowest		Control	Highest		Lowest		Control	Highest		Lowest		Control
% crude protein														
Parathion	23.09	Trichlorfon	22.07	22.15	Phosphamidon	23.47	Mevinphos	22.57	23.71	Parathion	25.99	Phosphamidon	25.14	25.42
% crude fiber														
Trichlorfon	25.19	Dimethoate	22.99	24.34	S D 7438	25.60	Phosphamidon	23.99	24.47	Dimethoate	24.12	S D 7438	23.35	23.69
% phosphorus														
Mevinphos	0.316	Trichlorfon	0.292	0.298	Diazinon	0.293	Azinphosmethyl	0.264	0.291	Azinphosmethyl	0.279	Malathion	0.257	0.271
% ether extract														
Azinphosmethyl	2.09	Trichlorfon	1.90	1.99	Malathion	1.38	S D 7438	1.22	1.25	Malathion	1.51	Phosphamidon	1.38	1.43
% nitrogen-free extract														
Dimethoate	38.64	Demeton	37.51	38.32	Azinphosmethyl	37.15	Diazinon	35.99	35.94	Phosphamidon	34.93	Parathion	33.49	34.61
% ash														
Demeton	9.94	Phosphamidon	8.86	9.11	Mevinphos	10.07	Parathion	9.37	9.83	Dimethoate	10.32	Phosphamidon	9.75	10.08

Table 15. Analysis of variance of percentage of crude protein in alfalfa sampled at various dates after application of 12 organophosphorus insecticides during summer growth of 1965 and spring and summer growth of 1966.

Source of variation	1965 Summer growth					1966 Spring growth					1966 Summer growth				
	df	SS	Mean S	F value Observed	F value Req'd*	df	SS	Mean S	F value Observed	F value Req'd*	df	SS	Mean S	F value Observed	F value Req'd*
Blocks	2	32.09				2	21.62				2	24.55	12.27		
Insecticides	12	16.15	1.35	0.82	2.18	9	13.34	1.48	0.82	2.46	12	11.57	0.96	1.21	2.18
Error (A)	24	39.57	1.65			18	30.51	1.69			24	19.17	0.80		
Time	4	382.06	95.51	108.76	2.46	4	306.44	76.61	102.61	2.48	5	989.00	197.80	285.22	2.21
Insec X Time	48	42.56	0.89	1.01	1.50	36	29.36	0.82	1.09	1.57	60	38.01	0.63	0.91	1.32
Error (B)	104	91.32	0.88			80	59.73	0.75			130	90.15	0.69		
Total	194	608.77				149	461.02				233	1172.4			

*at the .05 level of significance.

Table 16. Analysis of variance of percentage of crude fiber in alfalfa sampled at various dates after application of 12 organophosphorus insecticides during summer growth of 1965 and spring and summer growth of 1966.

Source of variation	1965 Summer growth					1966 Spring growth					1966 Summer growth				
	df	SS	Mean S	F value Observed	F value Req'd*	df	SS	Mean S	F value Observed	F value Req'd*	df	SS	Mean S	F value[a] Observed	F value[a] Req'd*
Blocks	2	11.31				2	6.03				2	17.12			
Insecticides	12	45.23	3.77	0.94	2.18	9	34.93	3.88	1.97	2.46	12	9.97	0.83	0.80	1.75
Error (A)	24	95.83	3.99			18	35.54	1.97			24	24.38	1.02		
Time	4	641.81	160.45	98.39	2.46	4	73.28	18.32	18.88	2.48	5	418.33	83.67	80.62	2.21
Insec X Time	48	77.88	1.62	0.99	1.50	36	35.45	0.98	1.02	1.57	60	72.26	1.20	1.16	1.32
Error (B)	104	169.60	1.63			80	77.62	0.97			130	135.45	1.04		
Total	194	1041.65				149	262.85				233	677.52			

[a]F value calculated with new error mean square = 1.037; error (A) SS and error (B) SS pooled and divided by pooled degrees of freedom for new error mean square since error (A) mean square < error (B) mean square.

* at the 0.05 level of significance.

Table 17. Analysis of variance of percentage of ether extract in alfalfa sampled at various dates after application of 12 organophosphorus insecticides during summer growth of 1965 and spring and summer growth of 1966.

Source of variation	1965 Summer growth					1966 Spring growth					1966 Summer growth				
	df	SS	Mean S	F value[a] Observed	Req'd*	df	SS	Mean S	F value Observed	Req'd*	df	SS	Mean S	F value[b] Observed	Req'd*
Blocks	2	0.20				2	0.05				2	0.30			
Insecticides	12	0.57	0.05	0.93	1.83	9	0.34	0.04	1.64	2.46	12	0.29	0.02	1.29	1.75
Error (A)	24	1.03	0.04			18	0.41	0.02			24	0.33	0.01		
Time	4	2.61	0.65	12.72	2.44	4	2.52	0.63	33.38	2.48	5	0.98	0.19	10.33	2.21
Insec X Time	48	1.55	0.03	0.63	1.46	36	0.38	0.01	0.56	1.57	60	1.19	0.02	1.06	1.32
Error (B)	104	5.52	0.05			80	1.51	0.02			130	2.57	0.02		
Total	194	11.48				149	5.22				233	5.66			

[a]F value calculated with new error mean square = 0.051; error (A) SS and error (B) SS pooled and divided by pooled degrees of freedom for new error mean square since error (A) mean square < error (B) mean square.

[b]F value calculated with new error mean square = 0.018.

* at the .05 level of significance.

Table 18. Analysis of variance of percentage of nitrogen-free extract sampled at various dates after application of 12 organophosphorus insecticides during summer growth of 1965 and spring and summer growth of 1966.

Source of variation	1965 Summer growth					1966 Spring growth					1966 Summer growth				
	df	SS	Mean S	F value Observed	Req'd*	df	SS	Mean S	F value Observed	Req'd*	df	SS	Mean S	F value[a] Observed	Req'd*
Blocks	2	12.70				2	14.70				2	46.92	23.46		
Insecticides	12	25.55	2.13	1.26	2.18	9	25.62	2.85	0.75	2.46	12	37.22	3.10	1.21	1.75
Error (A)	24	40.51	1.69			18	67.87	3.77			24	55.01	2.29		
Time	4	285.34	71.34	51.33	2.48	4	304.98	76.25	21.35	2.48	5	212.01	424.18	165.44	2.21
Insec X Time	48	33.85	0.71	0.51	1.50	36	117.94	3.28	0.92	1.57	60	159.39	2.65	1.04	1.32
Error (B)	104	144.54	1.39			80	285.65	3.57			130	339.85	2.61		
Total	194	542.49				149	816.76				233	275.93			

[a]F value calculated with new error mean square = 2.564; error (A) SS and error (B) SS pooled and divided by pooled degrees of freedom for new error mean square since error (A) mean square < error (B) mean square.

* at the .05 level of significance.

Table 19. Analysis of variance of percentage of phosphorus in alfalfa sampled at various dates after application of 12 organophosphorus insecticides during summer growth of 1965 and spring and summer growth of 1966.

Source of variation	1965 Summer growth					1966 Spring growth					1966 Summer growth				
	df	SS	Mean S	F value Observed	F value Req'd*	df	SS	Mean S	F value Observed	F value Req'd*	df	SS	Mean S	F value Observed	F value Req'd*
Blocks	2	0.0132				2	0.0036				2	0.0057			
Insecticides	12	0.0125	0.0010	1.86	2.18	9	0.0147	0.0016	0.81	2.46	12	0.0089	0.0007	0.50	2.18
Error (A)	24	0.0134	0.0006			18	0.0366	0.0020			24	0.0356	0.0015		
Time	4	0.2662	0.0665	449.87	2.46	4	0.0526	0.0132	49.25	2.48	5	0.6166	0.1233	114.36	2.21
Insec X Time	48	0.0093	0.0002	1.31	1.50	36	0.0088	0.0002	0.92	1.57	60	0.0817	0.0014	1.26	1.32
Error (B)	104	0.0153	0.0001			80	0.0214	0.0003			130	0.1401	0.0011		
Total	194	0.3302				149	0.1378				233	0.8889			

* at the .05 level of significance.

Table 20. Analysis of variance of percentage of ash in alfalfa sampled at various dates after application of 12 organophosphorus insecticides during summer growth of 1965 and spring and summer growth of 1966.

Source of variation	1965 Summer growth					1966 Spring growth					1966 Summer growth				
	df	SS	Mean S	F value Observed	F value Req'd*	df	SS	Mean S	F value Observed	F value Req'd*	df	SS	Mean S	F value[a] Observed	F value[a] Req'd*
Blocks	2	3.05				2	4.19				2	2.60			
Insecticides	12	12.90	1.07	0.65	2.18	9	6.51	0.72	0.46	2.46	12	5.86	0.49	0.52	1.75
Error (A)	24	39.93	1.66			18	28.61	1.58			24	20.99	0.87		
Time	4	249.26	62.31	110.11	2.46	4	90.86	22.72	35.63	2.48	5	105.55	21.11	22.49	2.21
Insec X Time	48	24.23	0.50	0.89	1.50	36	20.34	0.57	0.89	1.57	60	35.36	0.58	0.63	1.32
Error (B)	104	58.85	0.57			80	50.99	0.64			130	123.59	0.95		
Total	194	388.22				149	201.52				233	293.95			

[a]F value calculated with new error mean square = 0.94; error (A) SS and error (B) SS pooled and divided by pooled degrees of freedom for new error mean square since error (A) mean square < error (B) mean square.

* at the .05 level of significance.

Because of their increasing threat to world food and fiber supplies, interest in the study of insects has resulted in considerable growth in entomology as a career field.

Entomologists pursue research in colleges and universities; work in Federal, state and local health services as advisers to regulatory and government agencies; and are employed in private industry.

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